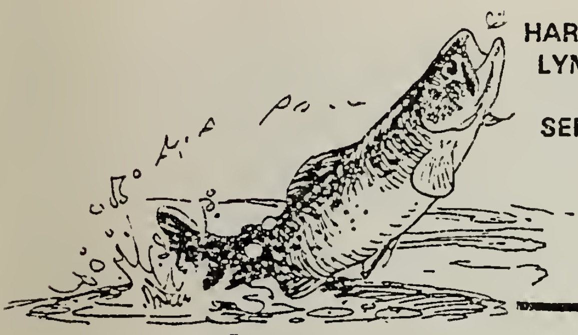
S
577.68
H2casr
1995

Bergman, Harold L
Comments on
ARCO's studies of
riparian mammals
on the upper Clark
Fork River and
Supplemental state

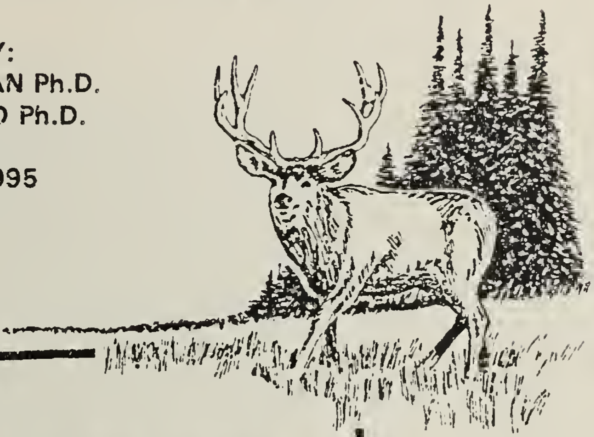
STATE OF MONTANA
L RESOURCE DAMAGE PROGRAM

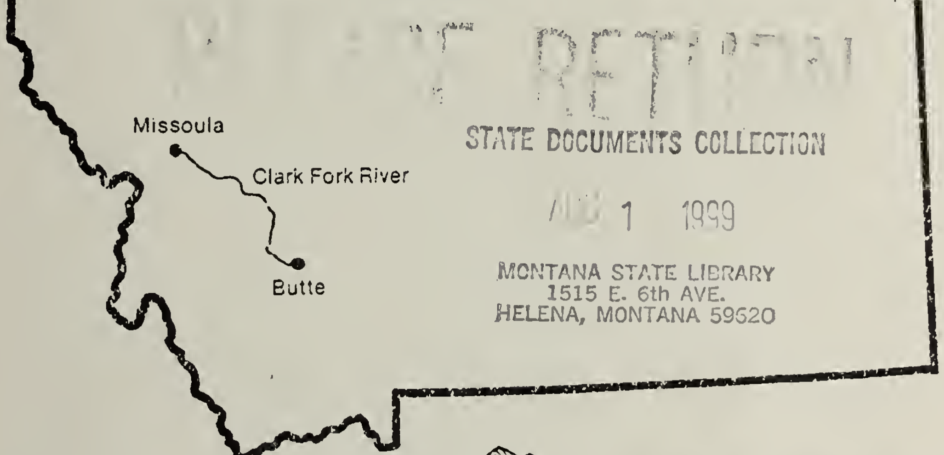
COMMENTS ON ARCO'S STUDIES OF RIPARIAN MAMMALS ON THE UPPER CLARK FORK RIVER
AND
SUPPLEMENTAL STATE REPORT
CONTINUED STUDIES ON THE EXPOSURE AND INJURY FROM ENVIRONMENTAL CONTAMINATION ON SEMI-AQUATIC MAMMALS IN THE UPPER CLARK FORK RIVER, MONTANA



PREPARED BY:
HAROLD L. BERGMAN Ph.D.
LYMAN McDONALD Ph.D.

SEPTEMBER 23, 1995





PLEASE RETURN
STATE DOCUMENTS COLLECTION
AUG 1 1999
MONTANA STATE LIBRARY
1515 E. 6th AVE.
HELENA, MONTANA 59620

MONTANA STATE LIBRARY
3 0864 0014 4529 8

S
577.68
H2casr
1995

Bergman, Harold L
Comments on ARCO's studies of riparian mammals on the upper Clark Fork River and Supplemental state

DATE DUE

MONTANA STATE LIBRARY
1515 East 6th Avenue
Helena, MT 59620-1800

AUG 11 1999

DEMCO

COMMENTS ON:

ARCO'S STUDIES OF RIPARIAN MAMMALS ON THE UPPER CLARK FORK RIVER

and

SUPPLEMENTAL STATE REPORT
Bergman, H.L. and M.J. Szumski. 1995.
Continued Studies on the Exposure and Injury from Environmental
Metal Contamination on Semi-Aquatic Mammals
in the Upper Clark Fork River, Montana.

Prepared by:

Harold L. Bergman

and

Lyman McDonald

Testifying Expert:

Harold L Bergman

Harold L. Bergman, Ph.D.
Department of Zoology and Physiology
University of Wyoming
Laramie, Wyoming 82071

and

Testifying Expert on Statistical and Sampling Issues:

Lyman McDonald

Lyman McDonald, Ph.D.
Western Ecosystems Technology, Inc.
Cheyenne, Wyoming 82001

Date:

September 23, 1995

General Comments

Overall, ARCO studies pertaining to the semi-aquatic mammals were haphazard, conducted in a statistically (and often times scientifically) indefensible manner, and presented in a way that can only be perceived as intentionally obscure. The apparent lack of protocols and standard operating procedures for all aspects of the ARCO studies raises further suspicions as to the quality of the data and bias of those who collected and interpreted the data.

The ARCO report is filled with inconsistencies that must be interpreted as post-hoc rationalization. During their riparian mammal surveys, ARCO consultants employed similar methods for quantifying mammal sign, and conducted their study on the same reference river, as used by the State. Data presented in ARCO's field data sheets unequivocally demonstrated reduced abundance of sign for three species of piscivorous mammal (river otter, mink, and raccoon) on the Clark Fork River compared to their own selected reference river (Table 1, attached). The ARCO report, however, conveniently concludes that both the State's (and their own) methods for determining animal abundance for mink and raccoon were invalid. Curiously, ARCO consultants then use the data collected during their "invalid" studies to conclude that there is no difference in species abundance, and that populations on the Clark Fork are healthy. A diagram of this illogical flow of logic contained in the ARCO report is presented in Figure 1. The only apparent explanation to this seemingly illogical progression is that ARCO consultants must have agreed with the validity of both the procedures and reference area selected by the State before their own data was collected. It was only after ARCO consultants conducted their own sign surveys (and obtained results similar to those of the State), that they then concluded their methods were invalid.

In a similar example of questionable logic, ARCO experts both defend the position that there is no reduction in abundance of semi-aquatic mammals on the Clark Fork, and conclude that the reason for significant differences in species abundance found in the State studies was not due to present mining impacts, but, rather, other human-related disturbances.

Specific Comments

(This section lists only the most serious flaws/discrepancies found in ARCO's methods, data and conclusions, and is by no means exhaustive.)

Training of field biologists in sign identification

State investigators were initially trained by state biologists/field researchers skilled in sign identification/discrimination techniques of the semi-aquatic species studied (otter, mink, raccoon, and beaver). The State's field biologists concurrently conducted field studies of otter, mink, raccoon, and beaver in Wyoming, Idaho, and Colorado, and have trained other field biologists in proper sign identification and discrimination for these as well as other species. Based on the resumes provided, ARCO's consultants had no prior experience concerning field research on these species, particularly concerning sign identification, and apparently received no field training a-priori. These researchers then appointed field technicians, again with no apparent prior training, to actually conduct the sign studies. Their lack of knowledge in proper sign identification is evident in the field data sheets provided by ARCO (e.g., LHW000000432) where there is clear uncertainty as to the identification of what could have been an otter latrine. Otter latrines are extremely unique in their appearance, with any confusion in their proper identification a clear sign of improper or no training.

Comparison of relative effort in the ARCO and State field studies

It should be noted that ARCO studies were conducted over a single field season, with approximately three weeks of field studies along the Clark Fork and Big Hole Rivers. Conclusions drawn from State studies were formulated after two field seasons of research with over 16 weeks of actual time spent in the field on the Clark Fork and Big Hole Rivers. Importantly, State studies and observations were conducted throughout the year, thereby allowing State investigators to base their conclusions regarding species abundance/habitat-disturbance considerations upon a much more inclusive and, therefore, more accurate base of information.

Problems in quality/completeness of documentation of ARCO riparian mammal surveys

Documentation of ARCO's methods employed for field collection and data analysis is incomplete. For example:

- No Standard Operating Procedures or Protocols were provided, or appear to exist.

- The methods listed on page 13 of the ARCO report are very brief and of too little detail to ascertain how data were actually collected. Based on the information provided, we can only conjecture approximately what was done, because what has been provided is incomplete.

- Much of the information we have received appears to be incomplete. For example, document LHW00001824 contains a SAS command file for making comparisons of sign between the two rivers by habitat type. The file has BEAVER and MKRAT crossed out as variables in the analysis. The subsequent pages only include results for MINK and COON, although if the

command file on this first page was run, BEAVER and MKRAT results would have been available, but apparently have been withheld by ARCO.

- The computer files used in these analyses (e.g., LCFVSBH.TXT) that appear as the input data in the hard copies of SAS command files, were not provided. Therefore, no verification of analysis or quality control/quality assurance can be conducted.

- It appears that only two of the twelve habitat summary sheets (the "no grass" and "conifer" habitat), listing map locations (map segments), reach, length, and mink sign according to particular habitat type for the Big Hole and Clark Fork, were provided (LHW000001841-1847).

The ARCO report erroneously states that ARCO riparian mammal/habitat surveys were conducted on the same river reaches sampled by the State.

- The ARCO report indicates that their river reaches were selected to match those surveyed by the State. This in fact was not the case. The State surveyed *six* Clark Fork River reaches from the Warm Springs Ponds to, but not including the Milltown Reservoir. Sites within each of these reaches were then matched to *six* corresponding, ecologically similar reference sites based on important environmental covariables (see Figure 3 in Bergman and Szumski 1994, and UW SOP P.33). In contrast, ARCO consultants arbitrarily defined *five* reaches on the Clark Fork that included the marsh habitat of the Milltown Reservoir. To these they compared *four* arbitrarily defined reaches on the Big Hole River, with no reference site of similar marsh habitat selected for comparison with the Milltown Reservoir (Figures M-12, M-13 in the ARCO report).

- The State examined semi-aquatic mammal sign and habitat variables in the riparian habitat of the Clark Fork River. Inference regarding the effects of current metal contamination on the piscivorous mammals in the State's study is therefore to those populations inhabiting the river, and does not include the contiguous but unique marsh habitat of the Milltown Reservoir. Similarly, because data was not collected from an appropriate marsh reference area during the ARCO field studies, information collected on the Milltown Reservoir by ARCO has no basis for comparison, and can not be used when drawing conclusions about relative mammal abundance between the Clark Fork and Big Hole Rivers.

The definitions of experimental units (segments within reaches, and the reaches themselves) used by ARCO investigators are completely arbitrary and are not independent.

- In the analysis of sign per river mile, the "sample size" used in the statistical analysis is completely subjective. Based on the information provided, it appears that each river was arbitrarily divided into segments of unequal length as defined in Figures M-12 and M-13 of the ARCO report. It is not apparent what criteria, if any, was used as the basis for the boundaries of each segment, since reach delineations are not indicated on the field data sheets, nor do they appear to correspond to any other predetermined criteria (see e.g., LHW00000910). The "sample size" used in most of the comparisons is the number of these arbitrary segments. The sample size could arbitrarily be doubled by further dividing each reach into twice as many segments, post-hoc. These segments are not independent from one another, and represent a census in space along each reach of the Clark Fork and Big Hole Rivers. In this sense, only point estimates should be given because results are not based on a sample of units.

- The reach definition is also completely arbitrary. In fact, it appears that there were 10 floats (10 days) conducted on the upper Clark Fork River and 6 floats (6 days) conducted on the Big Hole. The entire river was therefore censused and not systematically sub-sampled as reported.

- ARCO's consultants compared sign/mile between the Clark Fork and Big Hole for each habitat type (eg., tree type 0, shrub type 1, etc.) within each reach. For example, sign per mile in reach 1 on the Clark Fork for tree type 0 was compared to sign per mile in reach 1 of the Big Hole for tree type 0. The analysis therefore, is based on the idea that reach 1 of the Clark Fork and reach 1 of the Big Hole have been matched based on certain physical characteristics. The protocol for matching is not referenced anywhere in the documents we have received. If the ARCO consultants were trying to follow a design similar to that used by the State, they clearly did not understand the State's concept of the paired design. As is clearly explained in the State's Standard Operating Procedures (UW SOP P.33 and UW SOP P.34), Clark Fork Sites were randomly chosen within each of the six river reaches. Reference sites were then matched to each of these Clark Fork sites based on a number of physical habitat characteristics. In contrast, the ARCO studies censused each river, then later divided each river into arbitrary units of roughly equal lengths. Even without considering the serious flaws in ARCO's definition of the experimental unit, this arbitrary matching of sites based on post-hoc subdivision of sampling units is completely inappropriate and invalidates any statistical comparison of sign abundance on the two rivers.

Field methods employed by ARCO consultants were highly subjective, unscientific, and highly biased.

- Spring riparian mammal surveys on Silver Bow Creek and the upper Clark Fork River were conducted in the absence of any reference site surveys, making the data useless for any comparative conclusions regarding species abundance.

- The riparian survey method of floating downstream until "preferred areas of habitat" are observed is completely subjective, contains no method to ensure equal search effort between rivers, and is likely biased. The information gathered using this technique is only adequate for indicating species presence, and is completely inadequate for use in any type of quantitative comparisons between rivers.

- No measures were taken to prevent the double counting of sign left by the same individual. The result is an artificially and disproportionately inflated number of sign reported. During the State's sign surveys, in order to minimize double counting, sign of the same species (a track and latrine for example) discovered within 25 meters of each other were assumed to have been left by the same individual, and was therefore counted as only one sign event. Applying this criterion to ARCO's data, all of the otter sign discovered on one of the islands in the Milltown reservoir would have been counted as a single sign event, instead of the inflated number reported in the ARCO studies.

ARCO consultants inappropriately used opportunistic observations of semi-aquatic mammals in their assessment of abundance.

Assuming that the providers of such information are competent in distinguishing sign, such information could only, at best, be used as an indication of presence at the time of observation. Such data could in no way be added (as it was in the ARCO studies) as valid supplemental data to the riparian mammal surveys for the assessment of otter abundance between the Clark Fork and Big Hole Rivers. This is true for several reasons:

- All of ARCO's opportunistic observations were collected during intensive investigations associated with the NRDA of the Clark Fork River. During these studies, no similar investigations were conducted along the Big Hole or any other similar reference areas. This is an obvious example of unequal survey effort. To use these observations as supplemental data in a study that purports equality in effort is a gross flaw both in logic and the scientific method. Numerous opportunistic observations of otter sign were observed on the Big Hole River (while none was observed on the Clark Fork) by State researchers during times other than the sign survey. This information was not included as valid supplemental data by the State in their quantitative, statistical comparison of sign on the Clark Fork and Big Hole Rivers.

- While individuals reporting such opportunistic observations often have honest intentions, they are often not adequately qualified to accurately identify the sign of these species.

- Opportunistic observations can not distinguish between established residents and transient individuals. For example, opportunistic observations of otter along the Clark Fork at the

confluences of Racetrack and Rock Creek, as well as other alleged observations between the Warm Springs Ponds and Deer Lodge, were most likely not those of established resident otters, since otter presence was not confirmed anywhere along the upper Clark Fork River during either State or subsequent ARCO riparian mammal surveys. Similarly, a reported observation of otter sign in the vicinity of Warm Springs Ponds included in the ARCO report was also never confirmed, despite extensive searches by trained, knowledgeable Montana Department of Fish, Wildlife, and Parks (MDFWP) personnel who work daily on the Warm Springs Ponds Game Management Area (Dave Dziak, MDFWP, pers. comm.).

ARCO consultants misinterpreted the State's statistical comparisons, and the hypotheses tested for both semi-aquatic mammal surveys and MDFWP trapping records.

It appears that the ARCO consultants did not understand the hypotheses tested by the State for both the sign survey data and the comparisons of otter trapped historically on the Clark Fork and reference rivers. For the sign comparisons, one-sided hypotheses were tested by the State using the one-sample randomization procedure. That is, the alternative to the hypothesis of equality is that there were fewer sign/river mile on the Clark Fork River than on the Big Hole River. The t-test p-values reported in table S-22 of ARCO's riparian mammals report are for two-sided hypotheses. The corresponding one-sided p-values, as well as the p-values of the State's randomization procedure are listed in the table below. These one-sided values from the tests performed by ARCO are in close agreement with the one-sided p-values reported by the State (Bergman and Szumski 1995, attached).

Test on State's 1992 sign data	One-sided P-values Otter	Mink	Raccoon
State's Randomization Procedure	0.062	0.047	0.016
ARCO's Paired T-test	0.096	0.055	0.002
ARCO's Two-sample T-test	-	0.051	0.005

The ARCO consultants made the same error in their assessment of MDFWP trapping records for otter. One-sided hypotheses were tested by the State using the one-sample randomization procedure on these records; but again, the t-test p-values reported in table S-23 of ARCO's riparian mammals report are for two-sided hypotheses. The corresponding one-sided p-values from both the State's and ARCO's analyses are listed in the table below, and again are in close agreement.

Test	P-values of Otter Trapped per mile - Clark Fork/Big Hole River
State's Randomization Procedure	0.0469
ARCO's Paired T-test	0.0570
ARCO's Two-sample T-test	0.0445

Additionally, the ARCO report states that the abundance of otter sign found during the State's semi-aquatic mammal studies were not statistically significant. This is absolutely false. During the 1992 field season, otter sign on the Big Hole River was significantly greater than on the

upper Clark Fork at P=0.0625. Otter sign remained significantly more abundant on the Big Hole compared to the Clark Fork River for both the 1993 (P=0.0312), and the combined 1992 and 1993 field season's comparisons (P=0.0312).

The ARCO report ignores the results of species presence/habitat studies found during their own riparian mammal study in their formation of opinions regarding semi-aquatic mammal populations on the Clark Fork River.

- In spite of the serious flaws in experimental design, and given that tests for statistical significance can not be applied to the ARCO data because census techniques were employed, point estimates from ARCO studies quite clearly demonstrate that otter sign was essentially non-existent on the upper Clark Fork River. No otter sign was found during ARCO studies on the upper Clark Fork from Warm Springs Ponds to the Milltown Reservoir; however, they did find otter sign at several locations on the Big Hole for an overall mean of 0.08 otter sign/mi (Table 1, attached). Therefore, to conclude from the data collected during the ARCO studies that otter sign is similar to or greater than that found on the Big Hole during the time period of the study, is unfounded.

- ARCO's spring 1994 riparian mammal survey along nearly the entire length of Silver Bow Creek indicates that no otter or raccoon sign was present, and that mink sign was found in only one location (ARCO report, Table R-32). This is not surprising since the creek has been shown to be devoid of any significant fish populations, and the result further corroborates the State's argument.

- ARCO 1994 studies quite clearly indicate less sign per river mile for both mink and raccoon (Table 1, attached) on the upper Clark Fork River compared to the Big Hole River. This is not surprising since both piscivorous species were also shown to be reduced on the Clark Fork relative to the Big Hole during the State's semi-aquatic mammal investigations (Bergman and Szumski 1994, 1995).

- From the limited census data collected by the ARCO consultants, absolutely no conclusions regarding the health of semi-aquatic mammalian populations can be drawn. ARCO's attempt to infer population health from a study involving a few observations of sign and several unconfirmed opportunistic observations is analogous to observing several cattle tracks in the mud and concluding that the herd that left them is healthy.

- The ARCO consultant's manipulation of their own "disturbance" data is also deceptive. An examination of ARCO's field data sheets indicate that a total of eight possible disturbance categories were recorded. Grazing by livestock was one of these classifications, and was recorded as present on 45 of 48 map locations (LHW000000425-426, LHW000000434-435) on reach 1 of the Big Hole River. Amazingly, ARCO consultants then conclude that "Almost 99 percent of the Big Hole is unaffected by any of the measured disturbances within this reach" (Page 14, ARCO report). This form of disturbance (as well as hay production) exists to varying degrees along the entirety of both river courses examined in ARCO's investigation. Through the habitat evaluation methods employed by the State, the riparian zone immediately adjacent to the Clark Fork was shown not to differ significantly from the Big Hole River in

terms of the habitat type proportions, cover availability, bank characteristics, or canopy cover (Bergman and Szumski 1995, Bergman and Szumski 1994).

- The report by ARCO's consultants fails to demonstrate that any differences between the Clark Fork and Big Hole Rivers, in proximity to roads or railroad grades, explains the absence of a viable otter population on the Clark Fork River. Research conducted on otter populations in Wyoming, Idaho, and Colorado indicate that otter are capable of tolerating high levels of boating traffic in relatively narrow bodies of water, and that bridges offer no impedance to travel (Bergman and Szumski 1995). Such disturbance is far greater than any produced by motor vehicle traffic adjacent to the upper Clark Fork River. The argument that disturbance by roads or bridges explains the reduced otter abundance on the Clark Fork as proposed by ARCO consultants is therefore highly doubtful, as is the argument that reduced otter abundance is the result of significant mortality by motor vehicle accidents. To the State's knowledge, **no** such mortalities have been reported to MDFWP authorities, nor has ARCO provided evidence that **any** such mortality has occurred for otter on the upper Clark Fork.

- The occurrence of otter sign on the Milltown Reservoir reported by ARCO consultants can not be used to argue that otter are as abundant on the upper Clark Fork as on the Big Hole. But this information is important as evidence in refuting ARCO's claim that the Milltown Dam significantly impedes immigration by otters into the upper Clark Fork. The occurrence of otter sign on the Milltown Reservoir clearly indicates that there is a seed population (probably immigrants from the Blackfoot River), which would be capable of further immigration up the Clark Fork. This is the most probable source of the occasional transients seen on the Clark

Fork, and demonstrates that no physical barriers exist to significantly impede immigration. Most important, however, are the questions raised by this discovery. If such a pool of otter indeed exists, why then hasn't a viable population of otter established itself further up the Clark Fork and its tributaries?

References

Bergman, H.L., and M.J. Szumski. 1995. Supplemental Report: Continued Studies on the Exposure and Injury from Environmental Metal Contamination on Semi-Aquatic Mammals in the Upper Clark Fork River, Montana. Report to the State of Montana. September, 1995.

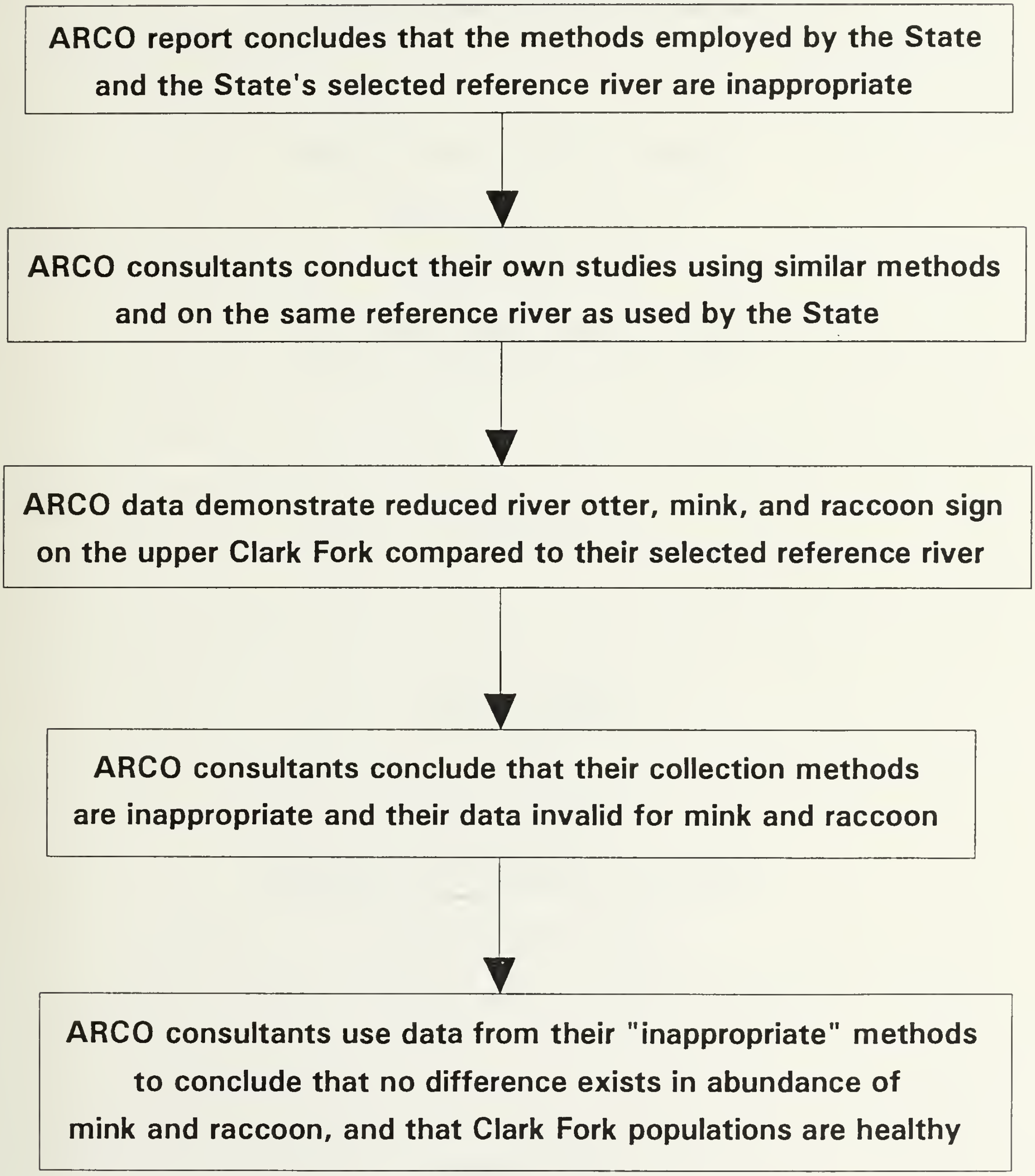
Bergman, H.L., and M.J. Szumski. 1994. Exposure to and injury from environmental metal contamination on semi-aquatic mammals in the upper Clark Fork River, Montana. Report to the State of Montana. December, 1994.

Dziak, D.J. Montana Department of Fish, Wildlife, and Parks. Warm Springs Ponds Game Management Area. Warm Springs, Montana.

Table 1. Overall results from fall 1994 ARCO riparian mammal surveys. Data are computed from observations reported in ARCO field data sheets.

	Clark Fork River	Big Hole River
Distance Surveyed (miles)	117.796	78.141
Total Otter Sign	0	6
Total Otter Sign per Mile	**0.00**	**0.08**
Total Mink Sign	31	137
Total Mink Sign per Mile	**0.26**	**1.75**
Total Raccoon Sign	5	47
Total Raccoon Sign per Mile	**0.04**	**0.60**

FIGURE 1. AN EXAMPLE OF THE QUESTIONABLE LOGIC CONTAINED IN ARCO'S RIPARIAN MAMMALS REPORT



ARCO report concludes that the methods employed by the State and the State's selected reference river are inappropriate

ARCO consultants conduct their own studies using similar methods and on the same reference river as used by the State

ARCO data demonstrate reduced river otter, mink, and raccoon sign on the upper Clark Fork compared to their selected reference river

ARCO consultants conclude that their collection methods are inappropriate and their data invalid for mink and raccoon

ARCO consultants use data from their "inappropriate" methods to conclude that no difference exists in abundance of mink and raccoon, and that Clark Fork populations are healthy

SUPPLEMENTAL REPORT

Continued Studies on the Exposure and Injury from Environmental Metal Contamination on Semi-Aquatic Mammals in the Upper Clark Fork River, Montana.

Research Report on Injury Determination

Wildlife Protocols #4 and #5

Assessment Plan, Part II

Clark Fork River Basin NPL Sites, Montana

Submitted by:

Harold L. Bergman

and

Michael J. Szumski

Red Buttes Environmental Biology Laboratory

University of Wyoming

Laramie, Wyoming 82071

September, 1995

INTRODUCTION

This report contains the findings from the second (1993) of two field seasons evaluating the continuing impact of past mining and smelting operations on piscivorous mammals (river otter *Lutra canadensis*, mink *Mustela vison*, and raccoon *Procyon lotor*) inhabiting the upper Clark Fork River in southwest Montana. This report integrates and presents conclusions based on both years' data. Findings from the first year's (1992) studies may be found in Bergman and Szumski (1994).

METHODS

The 1993 field season included the collection of additional mink from the upper Clark Fork River and reference streams. Animals were collected by the principal investigators, as well as collected from local trappers. Tissues from all mink were analyzed for metal residues, and those from fresh caught animals were also examined histologically, as before (Bergman and Szumski 1994). A second survey for semi-aquatic mammal sign, as well as a second habitat evaluation along paired Clark Fork and Big Hole River sites, were also conducted. Methods employed during the 1993 field season were identical to those of the 1992 season except where noted.

Mink Tissue Residue Analysis

Tissues (liver, kidney, and brain) were analyzed for Pb, Cd, As, Cu, and Zn as before (See UW SOP P.35, UW SOP P.36). Tissue metal residues were compared between Clark Fork and reference sites for the 1993 data and the combined 1992 and 1993 data for juveniles and adults by a two-sample, one-sided randomization test (Manly 1991).

Sign Survey for Semi-Aquatic Mammals

Sign surveys were conducted for river otter, mink, raccoon, and beaver employing identical methods, and on the same paired Clark Fork and reference sites as the 1992 surveys (See UW SOP P.33). The amount of sign per river mile observed in 1993 was compared between paired Clark Fork and Big Hole River sites using a one-sample randomization test. An overall analysis across both years' data was conducted by first averaging the differences between each reference and Clark Fork paired site obtained in 1992 and 1993, and then conducting a one-sample randomization test (Manly 1991) on these six averaged differences. A one-sided test was conducted on the piscivorous mammal sign (otter, mink, and raccoon), while a two-sided test was conducted on beaver sign.

Riparian Zone Habitat Analysis

As a complement to previous habitat studies that evaluated major habitat types on paired Clark Fork and reference areas, an evaluation of canopy cover was conducted on these same sites (See UW SOP P.34, P.34A). This evaluation focused on woody vegetation greater than 1 meter above ground, and included both shrubs and trees. After selecting a random starting point at each site, canopy coverage data was collected at five minute intervals along both sides of the river while walking the length of the entire reach. At each data collection point, transects were placed perpendicular to the shoreline, and extended inland 25 meters. The following distance intervals were demarcated on the transect: 0-1 m, 1-5 m, 5-25 m, and >25 m. These four distance intervals were given the distance category classification of 1, 2, 3, and 4 respectively. Beginning at the shoreline and proceeding in a consecutive manner to the center of each of the first three distance categories, observers recorded using ocular estimates, the category that first

contained greater than or equal to 75 percent canopy closure of woody vegetation. Category 4 was selected if none of the first three categories met the coverage criterion. Weights were assigned in decreasing magnitude to the four canopy cover transect classifications. Category 1,2,3, and 4 transects were assigned values of 4,3,2, and 1, respectively. After defining n_i $\{I=1,2,3,4\}$, as the number of transects classified as category I on each site, the index to canopy cover at each site, I, was calculated by:

$$I = \frac{4*n_1 + 3*n_2 + 2*n_3 + 1*n_4}{n_1 + n_2 + n_3 + n_4}$$

One-sample, two-sided randomization tests (Manly 1991) were conducted comparing the differences in proportion of category 1 classifications, and differences in the weighted cover indices I, between the reference and Clark Fork paired sites.

Mink Health/Age Assessment

Mink carcasses with tissues of suitable quality were examined grossly in the field and evaluated histologically by the Wyoming State Veterinary Pathologist, as before (See UW SOP P.32). Mink were aged in the same manner as before using canine tooth cementum annuli determined by the Wyoming Game and Fish Laboratory in Laramie, WY.

RESULTS

Mink Collection/Tissue Residue Analysis

Forty-three mink were collected during the 1993 trapping season, twenty-nine from the Clark Fork, and fourteen from reference streams. Twenty of the Clark Fork mink were male

(9 adults, 11 juveniles), and nine were female (5 adults, 4 juveniles). Of the mink collected from reference streams, ten were male (4 adults, 6 juveniles), and four were female (2 adults, 2 juveniles).

Tissue metal concentration means and standard errors of the means for liver, kidney, and brain from Clark Fork and reference site mink for 1993 and combined 1992 - 1993 data are found in Tables 1 through 6. Statistical test results (P-values) from 1993 and combined 1992 - 1993 comparisons between the Clark Fork and reference site mink tissue metal concentrations are found in Appendices 1 through 6.

Cadmium

In 1993, cadmium tissue residues were significantly elevated in adult livers from both the upper Clark Fork at P=0.0514, and combined Clark Fork Sites at P=0.0696 (Appendix 1). Combined 1992 - 1993 tissue comparisons indicated that cadmium was significantly elevated in adult livers collected from both the upper and combined Clark Fork River sites as compared to the reference sites (Appendix 4).

Lead

During 1993, lead was significantly elevated in the livers and kidneys of both juvenile and adult mink from both upper and combined Clark Fork River sites (Appendices 1 and 2), and was significantly elevated in brains of juveniles from the upper Clark Fork at P=0.0526 (Appendix 3).

Combined 1992 - 1993 comparisons were similar, demonstrating significantly elevated lead in the livers and kidneys of juveniles and adults in all Clark Fork site comparisons, as well

as significantly elevated in brains from adults from both upper and combined Clark Fork sites, and juveniles from the upper Clark Fork (Appendices 4 through 6). Additionally, lead residues in brains of juveniles from combined Clark Fork sites were significantly elevated at P=0.0542 (Appendix 6).

Copper

For 1993, copper residues were significantly elevated in adult liver and kidneys from both Clark Fork site comparisons, as well as adult brains from combined Clark Fork sites (Appendices 1 through 3). Copper was also significantly elevated in juvenile kidneys and brains from the upper Clark Fork sites (Appendices 2 and 3). Brains of adults from the upper Clark Fork, and juveniles from combined Clark Fork sites were significantly elevated at P=0.0640 and P=0.0505 respectively (Appendix 3).

Overall 1992 - 1993 data indicate that copper was significantly elevated in livers of adults and juveniles from the Clark Fork in all site comparisons (Appendix 4), as well as in kidneys from Clark Fork mink for all site/age comparisons except that of juveniles from the lower Clark Fork site (Appendix 5). Brains from adult mink from both Clark Fork site comparisons also had significantly elevated copper, as did juvenile brains from combined Clark Fork sites. Juvenile brains from the upper Clark Fork site were significantly elevated at P=0.0538 (Appendix 6).

Zinc

In 1993, zinc was significantly elevated in adult mink livers from both Clark Fork site comparisons (Appendix 1). Combined 1992 - 1993 data indicate that zinc was significantly

elevated in adult liver and kidneys from both upper and combined Clark Fork sites (Appendices 4 and 5).

Arsenic

For 1993, arsenic was significantly elevated in the livers of both adult and juvenile mink from the Clark Fork in all site comparisons (Appendix 1). Kidneys of juveniles from the upper Clark Fork site were also significantly elevated, and were significantly elevated in adults from this site at P=0.0616 (Appendix 2).

For 1992 - 1993 comparisons, arsenic was significantly elevated in both adult and juvenile livers and kidneys from the upper Clark Fork site, in juvenile livers from combined Clark Fork sites, and adult kidneys from combined Clark Fork sites (Appendices 4 and 5). Additionally, adult livers from combined Clark Fork sites were significantly elevated at P=0.0541 (Appendix 4).

Sign Survey for Semi-Aquatic Mammals

Significant differences were again found in the amount of semi-aquatic mammal sign between paired Clark Fork and reference sites (Tables 7 and 8). In 1993 otter and raccoon sign were significantly more abundant on reference sites than on paired Clark Fork sites, while mink sign was significantly more abundant at P=0.0625. Beaver sign was not significantly different between sites. In the overall analyses across data for both years, otter, mink, and raccoon sign were all significantly more abundant on reference sites than on paired Clark Fork sites, while beaver sign was not significantly different (Table 7 and 8).

Riparian Zone Habitat Analysis

There were no significant differences in the proportion of canopy cover 1 transects between paired Clark Fork and reference sites, nor was there a significant difference between sites in the weighted cover index.

Mink Health/Age Assessment

Of the forty-three mink collected, seventeen were suitable for histopathological evaluation. Of these, thirteen were from reference streams and four were from the upper Clark Fork River. Both reference and Clark Fork mink exhibited several common pathologies (mostly viral, fungal, and parasitic) similar to those seen the previous year, none of which were related to metal toxicity. The four Clark Fork mink demonstrated no overt signs of metal intoxication, however cellular changes observed in the body fat of one of the four (#211) was suggestive of possible increased fat peroxidation.

DISCUSSION

Results from the 1993 field season corroborate and strengthen those from the 1992 field season, and support our previous overall conclusions (Bergman and Szumski 1994) that populations of piscivorous mammals (river otter, mink, and raccoon) on the upper Clark Fork River and its tributaries continue to be negatively impacted by the effects of past mining and smelting operations.

Mink collected from the upper Clark Fork demonstrated significantly elevated tissue concentrations of the five mining-related metals (Pb, Cd, As, Cu, and Zn) elevated in the riparian zone. Tissue concentrations of mink caught on the upper Clark Fork were similar to those

reported in mink collected from other heavily-impacted, industrialized areas, while reference mink had concentrations similar to those reported in mink collected from non-industrialized areas (Blus and Henny 1990, Wren et al. 1988, Blus et al. 1987, Frank 1986, Ogle et al. 1985). Histopathologic studies of tissues taken from mink collected from the upper Clark Fork were inconclusive due to small sample size, however, tissues from several animals exhibited signs that were at least suggestive of metal-related effects. Even without overt signs of metal intoxication, metals could still be having a direct effect on the health of individual mink, and would be expected to have greater effects on species like the river otter, whose diet consists of a much greater proportion (nearly 100 percent) of aquatic prey items.

Sign surveys conducted over two field seasons along paired upper Clark Fork and reference sites on the Big Hole River conclusively show that populations of piscivorous mammals (otter, mink, and raccoon) are significantly reduced on the upper Clark Fork River. Importantly, herbivorous species such as beaver show no reduction on the upper Clark Fork, and are at levels comparable to that observed on reference sites. Species such as beaver *would* be expected to show a similar reduction in numbers as the piscivorous mammals if population declines were due to an overall reduction in habitat quality or an increase in human disturbance. Conversely, beaver *would not* be expected to show a similar population reduction as the piscivorous mammals if the ultimate cause was a metal-related effect, either from direct toxic effects due to the ingestion of contaminated aquatic food items, or due to indirect toxic effects from a reduced prey base. These findings corroborate prior analyses of MDFWP otter and beaver trapping records (Bergman and Szumski 1994), which indicated that significantly fewer otter are caught on the upper Clark Fork compared to both the Big Hole and Bitterroot Rivers. Importantly, no differences exist in the number of beaver caught within the MDFWP regions that contain these

rivers. River otter and beaver are considered to be closely associated species, with beaver improving the riparian habitat for otter through the improvement of fish habitat and the creation of bank dens, which are readily used as resting/escape cover and maternal dens by otters (Dubuc et al. 1990, Melquist and Dronkert 1987, Novak 1987, Mequist and Hornocker 1983). Because of this, otter are usually captured opportunistically by beaver trappers (H. Hash, MDFWP, pers. comm.). Therefore, two areas with similar beaver harvests should also have similar otter harvests if the otter populations are roughly equal. Both historical trapping records and our recent investigations clearly indicate that a viable otter population does not exist on the upper Clark Fork River, and that populations of two other piscivorous mammals on the upper Clark Fork (mink and raccoon) also are significantly reduced.

Assessments of habitat quality conducted on the upper Clark Fork and reference sites also indicate that reductions in populations of piscivorous mammals on the Clark Fork are due to a metal-related effect, and not due to differences in habitat characteristics or levels of human disturbance. Previous studies (Bergman and Szumski 1994) demonstrated that the upper Clark Fork does not differ significantly from the reference site in several habitat variables important to populations of piscivorous mammals including overall proportions of major habitat types, distance to nearest cover, proportions of nearest cover types, and bank height. Further habitat studies reported here demonstrate that canopy cover also does not differ significantly between the upper Clark Fork and reference sites.

Prior research (Bergman and Szumski 1994) also demonstrated that the upper Clark Fork was intermediate in disturbance levels when compared to two adjacent river drainages that both contain otter populations (the Big Hole and Bitterroot Rivers). The upper Clark Fork was

intermediate to these river valleys in terms of human populations, livestock densities, and irrigation demands.

Research conducted on river otter in both Wyoming and Idaho indicate that otter are capable of tolerating high levels of boating traffic in relatively narrow bodies of water. In these instances, otter became largely crepuscular or nocturnal, thereby avoiding the majority of the disturbance. Such disturbance by boat traffic is far greater than that produced by motor vehicle traffic adjacent to the upper Clark Fork River. River otter in Idaho and Wyoming were also observed to shift their activity spatially to avoid other areas of human disturbance such as towns and campgrounds. Bridges appeared to offer no impedance to travel (M. Szumski, unpublished observation). If an otter population could survive in the upper Clark Fork, it is likely that individuals would use similar temporal and spatial shifts in activity to avoid the majority of the motor vehicle and other human disturbance.

Findings by other researchers (Chapman 1993) indicate that trout populations are significantly reduced on the upper Clark Fork River. Trout can make up a significant proportion of both the otter and mink's diet (Bergman and Szumski 1994, Zackheim 1982). While Chapman (1993) did not quantify other species of fish important to piscivorous mammals, it is likely that they are also reduced on the upper Clark Fork River. Such a reduction in the aquatic food base would undoubtedly result in a reduced abundance of piscivorous mammals.

Trout, whitefish, and sucker collected from the upper Clark Fork had significantly higher whole-body concentrations of Cd, Pb, and Cu (Bergman and Szumski 1994, Bergman 1993). Applying these tissues concentrations to a tissue residue criterion model indicated that otter consuming fish from the upper Clark Fork River would exceed safe dietary levels of Pb, as would mink consuming a diet of greater than 65 percent fish (as suckers) (Bergman and Szumski

1994). Such information adds further evidence that direct toxic effects of metals on piscivorous mammals inhabiting the upper Clark Fork are possible, and may offer at least a partial explanation for the observed reduction in abundance of these species on the Clark Fork River.

CONCLUSIONS

The incorporation of a second year's field research further strengthens the conclusions drawn during the first year of study (Bergman and Szumski 1994). They are repeated below.

1. A viable otter population is absent from the upper Clark Fork River, based on complete absence of sign, information from MDFWP and BLM personnel, as well as local trappers and ranchers, and historical trapping records for otter, while they are present on all comparable rivers in western Montana.

2. Otter, mink, and raccoon abundance are significantly reduced on the upper Clark Fork River compared to paired reference sites.

3. Habitat structural features, which are important for otter and other piscivorous mammals in the riparian zone along the upper Clark Fork River, are identical (indistinguishable statistically) to the habitat structural features on paired reference sites on the Big Hole River, where otter, mink, and raccoons are more abundant.

4. The levels of human disturbance on otter and other piscivorous mammals caused by trapping, grazing, irrigation demands, proximity to highways and human populations appear to be similar or less severe in the upper Clark Fork valley as compared to similar river valleys in western Montana that currently contain otter.

5. Reductions in fish populations due to metals (Chapman 1993) have reduced the prey base for all piscivorous mammals on the upper Clark Fork River, with otter being the most severely affected due to their nearly complete reliance on a diet of fish.

6. Whole body concentrations of Pb, Cd, and Cu are significantly elevated in fish from the upper Clark Fork River as compared to fish from reference sites.

7. Metal residue concentrations for Pb, and sometimes Cd, Cu, Zn and As are significantly elevated in liver, kidney, and brain tissues from mink trapped on the upper Clark Fork River as compared to tissues from mink trapped from reference sites, and are comparable to elevated metal residues in tissues of mink collected in North America habitats contaminated with anthropogenic metal inputs.

8. Tissue histology suggests the possibility that metal-related cellular injury occurs to kidney proximal tubule epithelium.

9. Dietary exposure concentrations for Pb in mink consuming fish from the upper Clark Fork River, exceed the Pb tissue residue criterion (a safe dietary intake concentration of Pb), assuming that mink consume more than about 65 percent of their diet as suckers from below Warm Springs Ponds.

10. Dietary exposure concentrations for Pb in otter consuming fish from the upper Clark Fork River exceed the safe Pb tissue residue criterion, based on an estimate of probable exposure from consumption of 50 percent suckers and 50 percent trout from below Warm Springs Ponds.

11. Dietary exposure concentrations for Cd in the diets of mink and otter, though below the respective safe tissue residue criteria for Cd, may contribute to metal toxicity in mink and otter through synergistic interactions with Pb.

All of the above findings from this study and related studies, taken together in a reasonable weight-of-evidence analysis, support an overall conclusion that the observed absence of otter on the upper Clark Fork River is the direct result of past and present environmental metal contamination. Additionally, the findings from this study also support a conclusion that significantly reduced populations of mink and raccoon on the upper Clark Fork River are also directly related to past and present environmental metal contamination. For all piscivorous mammals in this study (otter, mink, and raccoon) the reduced prey base of fish populations on the upper Clark Fork River caused by metal contamination is probably the primary cause of this reduction with elevated tissue metal concentrations undoubtedly contributing to the overall effect. Furthermore, environmental metal contamination in the upper Clark Fork River has undoubtedly affected the ability of all tributaries within the upper Clark Fork River drainage to support otter, since the Clark Fork would be the main travel route between and into these areas. As water quality and fisheries within the upper Clark Fork improve, it is possible, but not inevitable that this river will once again be capable of supporting a viable otter population.

BIBLIOGRAPHY

Bergman, H.L. and M.J. Szumski. 1994. Exposure to and injury from environmental metal contamination on semi-aquatic mammals in the upper Clark Fork River, Montana. Report to the State of Montana, Natural Resource Damage Program. Helena, MT.

Bergman, H.L. 1993. The physiological impairment of fish caused by chronic exposure to metals at concentrations typically found in Clark Fork River food and water. Appendix F. in Aquatic Resources Injury Assessment Report, Upper Clark Fork River Basin. State of Montana, Natural Resource Damage Program. Helena, MT.

Blus, L.J., C.J. Henny, and B.M. Mulhern. 1987. Concentrations of metals in mink and other mammals from Washington and Idaho. Environ. Poll. 44:307-318.

Blus, L.J., and C.J. Henny. 1990. Lead and cadmium concentrations in mink from northern Idaho. Northwest Sci. 64:219-223.

Chapman, D. 1993. Assessment of injury to fish populations: Clark Fork River NPL Sites, Montana. Appendix G. in Aquatic Resources Injury Assessment Report, Upper Clark Fork River Basin. State of Montana, Natural Resource Damage Program. Helena, MT.

Dubuc, L.J., W.B. Krohn, and R.B. Owen, Jr. 1990. Predicting occurrence of river otters by habitat on Mount Desert Island, Maine. J. Wildl. Manage. 54(4):594-599.

Frank, A. 1986. In search of biomonitors for cadmium: cadmium content of wild swedish fauna during 1973-1976. Sci. Total Environ. 57:57-65.

Hash, H. Furbearer Biologist, Montana Department of Fish, Wildlife, and Parks. Missoula, Montana.

Manly, B.F.J. 1991. RT, A Program for Randomization Testing.

Melquist, W.E., and M.G. Hornocker. 1983. Ecology of river otters in west central Idaho. Wildl. Monogr. 83:1-60.

Melquist, W.E., and A. Dronkert. 1987. River Otter (Chapter 47). pp 627-641 In: Novak et al. (eds). Wild Furbearer Management and Conservation in North America. Ontario Trappers Assoc. Ontario, Canada.

Novak, M. 1987. Beaver (Chapter 25). pp 283-312 In: Novak et al. (eds). Wild Furbearer Management and Conservation in North America. Ontario Trappers Assoc. Ontario, Canada.

Ogle, M.C., P.F. Scanlon, R.L. Kirkpatrick, and J.V. Gwynn. 1985. Heavy metal concentrations in tissues of mink in Virginia. Bull. Environ. Contam. Toxicol. 35:29-37.

Wren, C.D., K.L. Fischer, and P.M. Stokes. 1988. Levels of lead, cadmium and other elements in mink and otter from Ontario, Canada. Environ. Poll. 52:193-202.

Zackheim, H.S. 1982. Ecology and population status of the river otter in southwestern Montana. M.S. thesis, University of Montana. Missoula, Montana. 100pp.

Table 1. Mink mean (and Standard Error of the Mean) liver metal concentrations by trapping location for mink collected on the Clark Fork River and reference sites during the 1993 trapping seasons.

		Mean (SEM)				
Location	N	Cadmium ug/g dw	Lead ug/g dw	Copper ug/g dw	Zinc ug/g dw	Arsenic ug/g dw
Juveniles						
Upper Clark Fork	12	0.5269	1.3417[a]	37.3461	97.0839	0.6890[a]
		(0.0841)	(0.2513)	(5.5724)	(8.2777)	(0.2110)
Lower Clark Fork[b]	3	0.2202	0.5344	38.1081	81.2613	-0.0313
		(0.0280)	(0.2131)	(11.0141)	(2.6089)	(0.0590)
Combined CFR Sites	15	0.4656	1.1802[a]	37.4985	93.9194	0.5449[a]
		(0.0745)	(0.2201)	(4.7949)	(6.7916)	(0.1844)
Combined Reference	8	0.7500	0.2744	28.0758	98.7597	0.1282
		(0.1840)	(0.1142)	(6.2480)	(7.9376)	(0.0939)
Adults						
Upper Clark Fork	12	1.1985	1.2093[a]	40.8664[a]	115.2527[a]	0.5594[a]
		(0.1333)	(0.2297)	(3.9603)	(5.3780)	(0.2564)
Lower Clark Fork[b]	2	1.1390	2.0750	74.4583	163.6595	0.1138
		(0.8508)	(1.5115)	(12.8466)	(61.8507)	(0.0726)
Combined CFR Sites	14	1.1900	1.3330[a]	45.6652[a]	122.1680[a]	0.4957[a]
		(0.1445)	(0.2654)	(4.8803)	(9.2242)	(0.2227)
Combined Reference	6	0.7904	0.2067	25.4963	94.7978	0.0310
		(0.2033)	(0.0466)	(4.4613)	(8.2487)	(0.0652)

[a] Indicates P ≤ 0.05 (see Appendix 1).

[b] No statistical comparison made due to small sample size.

Table 2. Mink mean (and Standard Error of the Mean) kidney metal concentrations by trapping location for mink collected on the Clark Fork River and reference sites during the 1993 trapping seasons.

		Mean (SEM)				
Location	N	Cadmium ug/g dw	Lead ug/g dw	Copper ug/g dw	Zinc ug/g dw	Arsenic ug/g dw
Juveniles						
Upper Clark Fork	12	0.9726 (0.0931)	1.7639[a] (0.3553)	25.6242[a] (2.2409)	80.7796 (5.4670)	0.7802[a] (0.3134)
Lower Clark Fork[b]	3	0.4879 (0.0942)	0.8872 (0.3205)	14.6258 (1.2495)	75.7601 (2.7846)	0.0327 (0.0169)
Combined CFR Sites	15	0.8757 (0.0916)	1.5886[a] (0.3018)	23.4245 (2.1409)	79.7757 (4.3928)	0.6307 (0.2610)
Combined Reference	8	1.7635 (0.4633)	0.3188 (0.1371)	19.6809 (1.9245)	102.6739 (9.4244)	0.2598 (0.0906)
Adults						
Upper Clark Fork	12	2.9948 (0.4498)	0.9539[a] (0.1581)	25.1789[a] (2.5474)	88.0235 (3.9145)	0.5915 (0.2489)
Lower Clark Fork[b]	2	2.0289 (1.4519)	1.5667 (0.5503)	34.4165 (2.4731)	113.5658 (22.0946)	-0.0206 (0.0095)
Combined CFR Sites	14	2.8568 (0.4227)	1.0414[a] (0.1581)	26.4986[a] (2.3617)	91.6724 (4.7564)	0.5041 (0.2201)
Combined Reference	6	3.3196 (1.4263)	0.1705 (0.0405)	13.6830 (0.6541)	80.5847 (4.5795)	0.0790 (0.1055)

[a] Indicates $P \leq 0.05$ (see Appendix 2).

[b] No statistical comparison made due to small sample size.

Table 3. Mink mean (and Standard Error of the Mean) brain metal concentrations by trapping location for mink collected on the Clark Fork River and reference sites during the 1993 trapping seasons.

		Mean (SEM)				
Location	N	Cadmium ug/g dw	Lead ug/g dw	Copper ug/g dw	Zinc ug/g dw	Arsenic ug/g dw
Juveniles						
Upper Clark Fork	12	0.0196 (0.0035)	0.2474 (0.0671)	12.4708[a] (0.3243)	60.5156 (0.9774)	0.0729 (0.0437)
Lower Clark Fork[b]	3	0.0630 (0.0448)	0.0454 (0.0079)	12.0170 (0.6949)	61.3005 (0.3721)	-0.0352 (0.0533)
Combined CFR Sites	15	0.0283 (0.0093)	0.2070 (0.0574)	12.3800 (0.2868)	60.6726 (0.7820)	0.0513 (0.0376)
Combined Reference	8	0.0243 (0.0051)	0.0999 (0.0494)	11.5904 (0.3502)	65.8558 (1.7133)	-0.0033 (0.0599)
Adults						
Upper Clark Fork	12	0.0445 (0.0078)	0.3483 (0.2446)	10.8726 (0.3645)	58.7121 (1.2769)	0.1142 (0.0674)
Lower Clark Fork[b]	2	0.0117 (0.0086)	0.0682 (0.0106)	11.5784 (1.1373)	53.7924 (3.3474)	-0.0143 (0.1318)
Combined CFR Sites	14	0.0398 (0.0074)	0.3083 (0.2101)	10.9734[a] (0.3395)	58.0093 (1.2384)	0.0958 (0.0604)
Combined Reference	6	0.0621 (0.0286)	0.0452 (0.0366)	9.7120 (0.6749)	62.5514 (3.0785)	0.0404 (0.0618)

[a] Indicates $P \leq 0.05$ (see Appendix 3).

[b] No statistical comparison made due to small sample size.

Table 4. Mink mean (and Standard Error of the Mean) liver metal concentrations by trapping location for mink collected on the Clark Fork River and reference sites during the combined 1992 and 1993 trapping seasons.

		Mean (SEM)				
Location	N	Cadmium ug/g dw	Lead ug/g dw	Copper ug/g dw	Zinc ug/g dw	Arsenic ug/g dw
Juveniles						
Upper Clark Fork	17	0.5673	1.1650[a]	42.4247[a]	102.1399	0.5618[a]
		(0.0873)	(0.1934)	(4.9100)	(6.4911)	(0.1561)
Lower Clark Fork	8	0.3072	0.5755[a]	41.3526[a]	85.7460	0.2420
		(0.0349)	(0.0802)	(3.9041)	(2.5637)	(0.1161)
Combined CFR Sites	25	0.4841	0.9763[a]	42.0816[a]	96.8939	0.4594[a]
		(0.0647)	(0.1439)	(3.5159)	(4.7065)	(0.1151)
Combined Reference	13	0.5674	0.2505	28.1625	94.9522	0.0883
		(0.1311)	(0.0740)	(3.8953)	(5.8703)	(0.0590)
Adults						
Upper Clark Fork	18	1.1583[a]	1.1283[a]	39.6863[a]	109.4792[a]	0.6868[a]
		(0.1027)	(0.1573)	(2.8747)	(4.6411)	(0.2814)
Lower Clark Fork[b]	2	1.1390	2.0750	74.4583	163.6595	0.1138
		(0.8508)	(1.5115)	(12.8466)	(61.8507)	(0.0726)
Combined CFR Sites	20	1.1564[a]	1.2230[a]	43.1635[a]	114.8972[a]	0.6295
		(0.1109)	(0.1903)	(3.6401)	(7.1683)	(0.2557)
Combined Reference	12	0.6120	0.1969	22.0716	89.9750	0.1500
		(0.1169)	(0.0547)	(2.6109)	(4.8274)	(0.0971)

[a] Indicates $P \leq 0.05$ (see Appendix 4).

[b] No statistical comparison made due to small sample size.

Table 5. Mink mean (and Standard Error of the Mean) kidney metal concentrations by trapping location for mink collected on the Clark Fork River and reference sites during the combined 1992 and 1993 trapping seasons.

		Mean (SEM)				
Location	N	Cadmium ug/g dw	Lead ug/g dw	Copper ug/g dw	Zinc ug/g dw	Arsenic ug/g dw
Juveniles						
Upper Clark Fork	17	1.0957 (0.1859)	1.4379[a] (0.2799)	24.9939[a] (1.6047)	82.6373 (4.2272)	0.7595[a] (0.2397)
Lower Clark Fork	8	0.5614 (0.0947)	0.7160[a] (0.1235)	16.5849 (1.0538)	75.3079 (2.0941)	0.1347 (0.0486)
Combined CFR Sites	25	0.9247 (0.1381)	1.2069[a] (0.2041)	22.3030[a] (1.3828)	80.2919 (2.9995)	0.5596 (0.1726)
Combined Reference	13	1.3029 (0.3279)	0.2933 (0.0878)	18.7849 (1.2815)	98.7832 (7.5647)	0.2652 (0.0811)
Adults						
Upper Clark Fork	18	2.9139 (0.3519)	0.9013[a] (0.1095)	22.7019[a] (1.9289)	84.5481[a] (3.0301)	0.7289[a] (0.2670)
Lower Clark Fork[b]	2	2.0289 (1.4519)	1.5667 (0.5503)	34.4165 (2.4731)	113.5658 (22.0946)	-0.0206 (0.0095)
Combined CFR Sites	20	2.8254 (0.3384)	0.9678[a] (0.1155)	23.8734[a] (1.9179)	87.4499[a] (3.7352)	0.6540[a] (0.2451)
Combined Reference	12	2.2170 (0.7727)	0.1906 (0.0577)	14.4020 (0.6028)	74.9852 (3.1871)	0.1297 (0.0732)

[a] Indicates $P \leq 0.05$ (see Appendix 5).

[b] No statistical comparison made due to small sample size.

Table 6. Mink mean (and Standard Error of the Mean) brain metal concentrations by trapping location for mink collected on the Clark Fork River and reference sites during the combined 1992 and 1993 trapping seasons.

		Mean (SEM)				
Location	N	Cadmium ug/g dw	Lead ug/g dw	Copper ug/g dw	Zinc ug/g dw	Arsenic ug/g dw
Juveniles						
Upper Clark Fork	17	0.0501 (0.0244)	0.2257[a] (0.0518)	14.1009 (0.7730)	61.3205 (1.8272)	0.1061 (0.0395)
Lower Clark Fork	8	0.0240 (0.0187)	0.0343 (0.0150)	13.6672 (0.6533)	54.5078 (2.5041)	0.0894 (0.0445)
Combined CFR Sites	25	0.0417 (0.0176)	0.1645 (0.0396)	13.9621[a] (0.5590)	59.1404 (1.5873)	0.1005 (0.0297)
Combined Reference	13	0.0164 (0.0046)	0.0688 (0.0321)	12.4266 (0.5701)	59.0450 (2.7619)	0.0373 (0.0462)
Adults						
Upper Clark Fork	18	0.0321 (0.0067)	0.2958[a] (0.1621)	11.0119[a] (0.3373)	54.6872 (1.6693)	0.2465 (0.1056)
Lower Clark Fork[b]	2	0.0117 (0.0086)	0.0682 (0.0106)	11.5784 (1.1373)	53.7924 (3.3474)	-0.0143 (0.1318)
Combined CFR Sites	20	0.0300 (0.0062)	0.2730[a] (0.1463)	11.0685[a] (0.3161)	54.5977 (1.5188)	0.2205 (0.0969)
Combined Reference	12	0.0518 (0.0167)	0.0516 (0.0199)	9.9087 (0.3822)	53.7765 (3.5281)	0.0893 (0.0390)

[a] Indicates $P \leq 0.05$ (see Appendix 6).

[b] No statistical comparison made due to small sample size.

Table 7. Mean sign per river mile of semi-aquatic mammals from 1993 and combined 1992-1993 sign surveys.

	1993 Mean Sign per River Mile			
Site	Otter	Mink	Raccoon	Beaver
Clark Fork River	0.00	1.55	3.26	11.77
Big Hole River	0.59[a]	2.86	7.50[a]	8.49

	1992 - 1993 Mean Sign per River Mile			
Site	Otter	Mink	Raccoon	Beaver
Clark Fork River	0.00	1.09	1.93	8.52
Big Hole River	0.52[a]	2.33[a]	5.02[a]	6.66

[a] Indicates $P \leq 0.05$ (see Table 8).

Table 8. Results of statistical comparisons (P values) of semi-aquatic mammal sign between the upper Clark Fork and Big Hole Rivers.

	P values			
Field Season	Otter	Mink	Raccoon	Beaver
1992	0.0625	0.0625	0.0156	0.5781
1993	0.0312	0.0625	0.0156	0.7812
1992 and 1993	0.0312	0.0469	0.0156	0.7344

Appendix 1. Statistical comparison of 1993 field season mink liver metal concentrations including P values of mean difference (≤ observed) and F ratio (largest variance /smallest variance ≥ observed) of a two sample randomized test. See Table 1 for metal concentration values.

Age/Site Comparison	P values Mean Difference (F ratio)				
	Cadmium	Lead	Copper	Zinc	Arsenic
Adult UCFR V REF	0.0514 (0.8593)	0.0001 (0.0001)	0.0120 (0.5598)	0.0234 (0.8612)	0.0126 (0.1222)
Juvenile UCFR V REF	0.8815 (0.1513)	0.0010 (0.0097)	0.1484 (0.9169)	0.5443 (0.4165)	0.0017 (0.4109)
Adult ACFR V REF	0.0696 (0.7779)	0.0001 (0.0002)	0.0070 (0.2807)	0.0170 (0.7173)	0.0166 (0.1869)
Juvenile ACFR V REF	0.9432 (0.1540)	0.0008 (0.0231)	0.1199 (0.9328)	0.6677 (0.5733)	0.0278 (0.4047)

UCFR = upper Clark Fork River sites, ACFR = all Clark Fork River sites combined, REF = reference sites.

Appendix 2. Statistical comparison of 1993 field season mink kidney metal concentrations including P values of mean difference (≤ observed) and F ratio (largest variance /smallest variance ≥ observed) of a two sample randomized test. See Table 2 for metal concentration values.

Age/Site Comparison	P values Mean Difference (F ratio)				
	Cadmium	Lead	Copper	Zinc	Arsenic
Adult UCFR V REF	0.6111 (0.2910)	0.0001 (0.0006)	0.0022 (0.0004)	0.1298 (0.6686)	0.0616 (0.2699)
Juvenile UCFR V REF	0.9666 (0.0880)	0.0002 (0.0033)	0.0314 (0.7607)	0.9738 (0.5047)	0.0349 (0.5152)
Adult ACFR V REF	0.6572 (0.2611)	0.0001 (0.0001)	0.0006 (0.0002)	0.0784 (0.5178)	0.0981 (0.2915)
Juvenile ACFR V REF	0.9860 (0.0911)	0.0002 (0.0071)	0.1254 (0.5831)	0.9890 (0.3365)	0.1197 (0.6111)

UCFR = upper Clark Fork River sites, ACFR = all Clark Fork River sites combined, REF = reference sites.

Appendix 3. Statistical comparison of 1993 field season mink brain metal concentrations including P values of mean difference (≤ observed) and F ratio (largest variance /smallest variance ≥ observed) of a two sample randomized test. See Table 3 for metal concentration values.

Age/Site Comparison	P values Mean Difference (F ratio)				
	Cadmium	Lead	Copper	Zinc	Arsenic
Adult UCFR V REF	0.7448 (0.4207)	0.0786 (0.6646)	0.0640 (0.6061)	0.9053 (0.1704)	0.2802 (0.7645)
Juvenile UCFR V REF	0.7728 (0.6825)	0.0526 (0.5147)	0.0448 (0.6234)	0.9957 (0.6466)	0.1447 (0.7330)
Adult ACFR V REF	0.8108 (0.4490)	0.0828 (0.7055)	0.0442 (0.6007)	0.9387 (0.2171)	0.3358 (0.7389)
Juvenile ACFR V REF	0.5321 (0.7959)	0.1077 (0.6000)	0.0505 (0.6370)	0.9986 (0.5171)	0.2129 (0.6455)

UCFR = upper Clark Fork River sites, ACFR = all Clark Fork River sites combined, REF = reference sites.

Appendix 4. Statistical comparison of 1992-93 field season mink liver metal concentrations including P values of mean difference (≤ observed) and F ratio (largest variance /smallest variance ≥ observed) of a two sample randomized test. See Table 4 for metal concentration values.

	P values Mean Difference (F ratio)				
Age/Site Comparison	Cadmium	Lead	Copper	Zinc	Arsenic
Adult UCFR V REF	0.0012 (0.7336)	0.0001 (0.0002)	0.0003 (0.2439)	0.0036 (0.6333)	0.0402 (0.3971)
Juvenile UCFR V REF	0.5088 (0.4309)	0.0001 (0.0040)	0.0190 (0.3236)	0.2170 (0.2920)	0.0004 (0.2942)
Juvenile LCFR V REF	0.9339 (0.0265)	0.0054 (0.6450)	0.0218 (0.6240)	0.8763 (0.0114)	0.0992 (0.2635)
Adult ACFR V REF	0.0011 (0.3319)	0.0001 (0.0001)	0.0001 (0.0531)	0.0019 (0.4170)	0.0541 (0.4086)
Juvenile ACFR V REF	0.7321 (0.3120)	0.0001 (0.0178)	0.0056 (0.4989)	0.4010 (0.6217)	0.0016 (0.2093)

UCFR = upper Clark Fork River sites, LCFR = lower Clark Fork River sites, ACFR = all Clark Fork River sites combined, REF = reference sites.

Appendix 5. Statistical comparison of 1992-93 field season mink kidney metal concentrations including P values of mean difference ($\leq$ observed) and F ratio (largest variance /smallest variance $\geq$ observed) of a two sample randomized test. See Table 5 for metal concentration values.

Age/Site Comparison	P values Mean Difference (F ratio)				
	Cadmium	Lead	Copper	Zinc	Arsenic
Adult UCFR V REF	0.1998 (0.3333)	0.0001 (0.0042)	0.0002 (0.0001)	0.0196 (0.6908)	0.0148 (0.1395)
Juvenile UCFR V REF	0.7247 (0.4006)	0.0001 (0.0021)	0.0019 (0.6923)	0.9710 (0.1083)	0.0142 (0.3613)
Juvenile LCFR V REF	0.9709 (0.0936)	0.0066 (0.7772)	0.8819 (0.4087)	0.9885 (0.0025)	0.8723 (0.0773)
Adult ACFR V REF	0.2250 (0.3155)	0.0001 (0.0012)	0.0002 (0.0003)	0.0105 (0.4461)	0.0285 (0.1414)
Juvenile ACFR V REF	0.8878 (0.3224)	0.0001 (0.0068)	0.0471 (0.5298)	0.9935 (0.0525)	0.1004 (0.3742)

UCFR = upper Clark Fork River sites, LCFR = lower Clark Fork River sites,
ACFR = all Clark Fork River sites combined, REF = reference sites.

Appendix 6. Statistical comparison of 1992-93 field season mink brain metal concentrations including P values of mean difference (≤ observed) and F ratio (largest variance /smallest variance ≥ observed) of a two sample randomized test. See Table 6 for metal concentration values.

Age/Site Comparison	P values Mean Difference (F ratio)				
	Cadmium	Lead	Copper	Zinc	Arsenic
Adult UCFR V REF	0.8776 (0.2752)	0.0055 (0.1308)	0.0217 (0.8179)	0.3976 (0.0355)	0.1178 (0.1262)
Juvenile UCFR V REF	0.1188 (0.3984)	0.0059 (0.2143)	0.0538 (0.2808)	0.2507 (0.5601)	0.1309 (0.9750)
Juvenile LCFR V REF	0.3504 (0.4737)	0.7766 (0.0540)	0.0971 (0.8241)	0.8537 (0.2192)	0.2171 (0.5497)
Adult ACFR V REF	0.9120 (0.2580)	0.0072 (0.2194)	0.0129 (0.8387)	0.4016 (0.0287)	0.1703 (0.1377)
Juvenile ACFR V REF	0.1759 (0.2236)	0.0542 (0.3566)	0.0417 (0.4158)	0.4954 (0.6088)	0.1206 (0.7699)

UCFR = upper Clark Fork River sites, LCFR = lower Clark Fork River sites, ACFR = all Clark Fork River sites combined, REF = reference sites.